

Restriction Digest of LT10 Promoter Mini-Prepped DNA

Made with Benchling

Project: Awesome Possum

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Purpose:

Digest Mini-prepped LT10 promoter DNA with restriction enzymes to prepare for gel electrophoresis separation of fragment construct from plasmid.

Single Digest:

- RFP in PSB mini-prepped DNA with EcoRI
- Colony 1 from 1st culture DNA with EcoRI

Mastermix:

- 30.0 μL dH₂O
- 4.0 μL Green fast digest buffer
- 2.0 μL Fast Digest EcoRI

Double Digest:

- Colonies 1, 2, 4, 7, 8, and 10 from first culture. Cut with EcoRI and PSTI
- Colonies 8, 9, and 14 from 2nd culture. Cut with EcoRI and PSTI.

Mastermix:

- 120.0 μL dH₂O
- 4.0 μL Fast Digest EcoRI
- 4.0 μL Fast Digest PstI
- 16.0 μL Green fast digest buffer

Both Single and double digest reaction tubes received 18.0 μL of corresponding mastermix. Next 2.0 μL of sample DNA was added to each tube. After that the tubes were gently flicked to mix and then placed into a hot water bath 37.0°C for 60.0 minutes.